<110> Watkins, Jeffry D.
      Huse, William D.
      Vasserot, Alain P.
      Marquis, David P.
      Smith, Eric P.

<120> Methods of Optimizing Antibody Variable Region Binding Affinity

<130> AME-08122

<140> 10/697,399
<141> 2003-10-30

<160> 50

<170> PatentIn version 3.2

<210> 1
<211> 107
<212> PRT
<213> Mus musculus

<400> 1

Asp Ile Val Leu Thr Gln Ser Pro Ala Thr Leu Ser Val Thr Pro Gly
1   5   10   15

Asp Arg Val Ser Leu Ser Cys Arg Ala Ser Gln Ser Ile Ser Asp Tyr
            20   25   30

Leu His Trp Tyr Gln Gln Lys Ser His Glu Ser Pro Arg Leu Leu Ile
            35   40   45

Lys Tyr Ala Ser His Ser Ile Ser Gly Ile Pro Ser Arg Phe Ser Gly
    50   55   60

Ser Gly Ser Gly Ser Asp Phe Thr Leu Ser Ile Asn Ser Val Glu Pro
65   70   75   80

Glu Asp Val Gly Ile Tyr Tyr Cys Gln His Gly His Ser Phe Pro Arg
                85   90   95

Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys
            100   105

<210> 2
<211> 107
<212> PRT
<213> Homo sapiens

<400> 2

Glu Ile Val Leu Thr Gln Ser Pro Ala Thr Leu Ser Leu Ser Pro Gly
1   5   10   15

Glu Arg Ala Thr Leu Ser Cys Arg Ala Ser Gln Ser Val Ser Ser Tyr
            20   25   30

Leu Ala Trp Tyr Gln Gln Lys Pro Gly Gln Ala Pro Arg Leu Leu Ile
        35   40   45

Tyr Asp Ala Ser Asn Arg Ala Thr Gly Ile Pro Ala Arg Phe Ser Gly
    50   55   60

Ser Gly Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Glu Pro
65   70   75   80

Glu Asp Phe Ala Val Tyr Tyr Cys Gln Gln Arg Ser Asn Trp Pro Leu
            85   90   95

Thr Phe Gly Gly Gly Thr Lys Val Glu Ile Lys
            100   105

<210> 3
<211> 122
<212> PRT
<213> Mus musculus

<400> 3

Gln Ile Gln Leu Val Gln Ser Gly Pro Glu Leu Lys Lys Pro Gly Glu
1   5   10   15

Thr Val Arg Ile Ser Cys Lys Ala Ser Gly Tyr Ala Phe Thr Thr Thr
            20   25   30

Gly Met Gln Trp Val Gln Glu Met Pro Gly Lys Gly Leu Lys Trp Ile
        35   40   45

Gly Trp Ile Asn Thr His Ser Gly Val Pro Lys Tyr Val Glu Asp Phe
    50   55   60

Lys Gly Arg Phe Ala Phe Ser Leu Glu Thr Ser Ala Asn Thr Ala Tyr
65   70   75   80

Leu Gln Ile Ser Asn Leu Lys Asn Glu Asp Thr Ala Thr Tyr Phe Cys
            85   90   95

Val Arg Ser Gly Asn Gly Asn Tyr Asp Leu Ala Tyr Phe Ala Tyr Trp
                100   105   110

Gly Gln Gly Thr Leu Val Thr Val Ser Ala
            115   120

<210> 4
<211> 113
<212> PRT
<213> Homo sapiens

<400> 4

Gln Val Gln Leu Val Gln Ser Gly Ser Glu Leu Lys Lys Pro Gly Ala
1   5   10   15

Ser Val Lys Val Ser Cys Lys Ala Ser Gly Tyr Thr Phe Thr Ser Tyr
                20   25   30

Ala Met Asn Trp Val Arg Gln Ala Pro Gly Gln Gly Leu Glu Trp Met
            35   40   45

Gly Trp Ile Asn Thr Asn Thr Gly Asn Pro Thr Tyr Ala Gln Gly Phe
        50   55   60

Thr Gly Arg Phe Val Phe Ser Leu Asp Thr Ser Val Ser Thr Ala Tyr
65   70   75   80

Leu Gln Ile Ser Ser Leu Lys Ala Glu Asp Thr Ala Val Tyr Tyr Cys
                85   90   95

Ala Arg Tyr Phe Asp Tyr Trp Gly Gln Gly Thr Leu Val Thr Val Ser
            100   105   110

Ser

<210> 5
<211> 120
<212> PRT
<213> Mus musculus

<400> 5

Val Gln Leu Leu Glu Ser Gly Pro Gly Leu Val Ala Pro Ser Gln Ser
1   5   10   15

Leu Ser Ile Thr Cys Thr Val Ser Gly Phe Ser Leu Thr Asp Tyr Gly
                20   25   30

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Val Asp Trp Val Arg Gln Pro Pro Gly Lys Gly Leu Glu Trp Leu Gly
        35   40   45

Met Ile Trp Gly Asp Gly Ser Thr Asp Tyr Asn Ser Ala Leu Lys Ser
        50   55   60

Arg Leu Ser Ile Thr Lys Asp Asn Ser Lys Ser Gln Val Phe Leu Lys
65   70   75   80

Met Asn Ser Leu Gln Thr Asp Asp Thr Ala Arg Tyr Tyr Cys Val Arg
                85   90   95

Asp Pro Ala Asp Tyr Gly Asn Tyr Asp Tyr Ala Leu Asp Tyr Trp Gly
            100   105   110

Gln Gly Thr Ser Val Thr Val Ser
        115   120

<210>   6
<211>   99
<212>   PRT
<213>   Mus musculus

<400>   6

Ser Ser Leu Ser Ala Ser Leu Gly Asp Arg Val Thr Ile Ser Cys Ser
1   5   10   15

Ala Ser Gln Asp Ile Asn Lys Tyr Leu Asn Trp Tyr Gln Gln Lys Pro
            20   25   30

Asp Gly Ala Val Lys Leu Leu Ile Phe Tyr Thr Ser Ser Leu His Ser
        35   40   45

Gly Val Pro Ser Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Tyr Ser
    50   55   60

Leu Thr Ile Ser Asn Leu Glu Pro Glu Asp Ile Ala Thr Tyr Tyr Cys
65   70   75   80

Gln Gln Tyr Glu Lys Leu Pro Trp Thr Phe Gly Gly Gly Thr Lys Leu
                85   90   95

Glu Val Lys

<210> 7
<211> 81
<212> PRT
<213> Homo sapiens

<400> 7

Glu Val Gln Leu Val Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly
1   5   10   15

Ser Leu Arg Leu Ser Cys Ala Ala Ser Trp Val Arg Gln Ala Pro Gly
            20   25   30

Lys Gly Leu Glu Trp Val Gly Arg Phe Thr Ile Ser Arg Asp Asp Ser
            35   40   45

Lys Asn Ser Leu Tyr Leu Gln Met Asn Ser Leu Lys Thr Glu Asp Thr
        50   55   60

Ala Val Tyr Tyr Cys Ala Arg Trp Gly Gln Gly Thr Thr Val Thr Val
65   70   75   80

Ser

<210> 8
<211> 80
<212> PRT
<213> Homo sapiens

<400> 8

Asp Ile Gln Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly
1   5   10   15

Asp Arg Val Thr Ile Thr Cys Trp Tyr Gln Gln Lys Pro Gly Lys Ala
            20   25   30

Pro Lys Leu Leu Ile Tyr Gly Val Pro Ser Arg Phe Ser Gly Ser Gly
            35   40   45

Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro Glu Asp
        50   55   60

Phe Ala Thr Tyr Tyr Cys Phe Gly Gly Gly Thr Lys Val Glu Ile Lys
65   70   75   80

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<210>    9
<211>    11
<212>    PRT
<213>    Artificial Sequence

<220>
<223>    Synthetic

<400>    9

Ser Ala Ser Gln Asp Ile Asn Asp Tyr Leu Asn
1                5                   10


<210>    10
<211>    33
<212>    PRT
<213>    Artificial Sequence

<220>
<223>    Synthetic

<400>    10

Ala Gly Thr Gly Cys Ala Ala Gly Thr Cys Ala Gly Gly Ala Cys Ala
1                5                   10                  15


Thr Thr Ala Ala Cys Gly Ala Cys Thr Ala Thr Thr Thr Ala Ala Ala
             20                  25                  30


Cys


<210>    11
<211>    7
<212>    PRT
<213>    Artificial Sequence

<220>
<223>    Synthetic

<400>    11

Gly Thr Ser Ser Leu His Ser
1                5


<210>    12
<211>    21
<212>    PRT
<213>    Artificial Sequence

<220>
<223>    Synthetic
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<400> 12

Gly Gly Cys Ala Cys Ala Thr Cys Ala Ala Gly Thr Thr Thr Ala Cys
1   5   10   15

Ala Cys Thr Cys Ala
          20

<210> 13
<211> 7
<212> PRT
<213> Artificial Sequence

<220>
<223> Synthetic

<400> 13

Asn Thr Ser Ser Leu His Ser
1   5

<210> 14
<211> 21
<212> PRT
<213> Artificial Sequence

<220>
<223> Synthetic

<400> 14

Ala Ala Cys Ala Cys Ala Thr Cys Ala Ala Gly Thr Thr Thr Ala Cys
1   5   10   15

Ala Cys Thr Cys Ala
          20

<210> 15
<211> 7
<212> PRT
<213> Artificial Sequence

<220>
<223> Synthetic

<400> 15

Tyr Thr Ser Val Leu His Ser
1   5

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<210>   16
<211>   21
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic

<400>   16

Thr Ala Cys Ala Cys Ala Thr Cys Ala Gly Thr Thr Thr Thr Ala Cys
1                   5                   10                  15


Ala Cys Thr Cys Ala
            20


<210>   17
<211>   7
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic

<400>   17

Asn Thr Ser Val Leu His Ser
1                   5


<210>   18
<211>   21
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic

<400>   18

Ala Ala Cys Ala Cys Ala Thr Cys Ala Gly Thr Thr Thr Thr Ala Cys
1                   5                   10                  15


Ala Cys Thr Cys Ala
            20


<210>   19
<211>   7
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic

<400>   19

Tyr Thr Ser Ser Leu His Val
1                   5
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```
<210>  20
<211>  21
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic

<400>  20

Thr Ala Cys Ala Cys Ala Thr Cys Ala Ala Gly Thr Thr Thr Ala Cys
1                   5                   10                  15


Ala Cys Gly Thr Gly
            20


<210>  21
<211>  7
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic

<400>  21

Asn Thr Ser Ser Leu His Val
1                   5


<210>  22
<211>  21
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic

<400>  22

Ala Ala Cys Ala Cys Ala Thr Cys Ala Ala Gly Thr Thr Thr Ala Cys
1                   5                   10                  15


Ala Cys Gly Thr Ala
            20


<210>  23
<211>  9
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic
```

```
<400>  23

Gln Gln Tyr Glu Asp Leu Pro Trp Thr
1               5


<210>  24
<211>  27
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic

<400>  24

Cys Ala Gly Cys Ala Gly Thr Ala Thr Gly Ala Ala Gly Ala Thr Cys
1               5                   10                  15


Thr Thr Cys Cys Gly Thr Gly Gly Ala Cys Gly
            20                  25


<210>  25
<211>  10
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic

<400>  25

Gly Phe Ser Leu Gly Asp Tyr Gly Val Asp
1               5                   10


<210>  26
<211>  30
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic

<400>  26

Gly Gly Ala Thr Thr Cys Thr Cys Ala Thr Thr Ala Gly Gly Cys Gly
1               5                   10                  15


Ala Cys Thr Ala Thr Gly Gly Thr Gly Thr Ala Gly Ala Cys
            20                  25                  30
```

```
<210>   27
<211>   8
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic

<400>   27

Met Ile Trp Pro Asp Gly Ser Thr
1                   5


<210>   28
<211>   24
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic

<400>   28

Ala Thr Gly Ala Thr Ala Thr Gly Gly Cys Cys Gly Gly Ala Thr Gly
1               5                   10                  15


Gly Ala Ala Gly Cys Ala Cys Ala
            20


<210>   29
<211>   8
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic

<400>   29

Met Ile Trp Gln Asp Gly Ser Thr
1                   5


<210>   30
<211>   24
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic

<400>   30

Ala Thr Gly Ala Thr Ala Thr Gly Gly Cys Ala Gly Gly Ala Thr Gly
1               5                   10                  15


Gly Ala Ala Gly Cys Ala Cys Ala
            20
```

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<210>   31
<211>   8
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic

<400>   31

Met Ile Trp Gly Asp Gly Ser Val
1               5


<210>   32
<211>   24
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic

<400>   32

Ala Thr Gly Ala Thr Ala Thr Gly Gly Gly Gly Thr Gly Ala Thr Gly
1               5                   10                  15


Gly Ala Ala Gly Cys Gly Thr Ala
            20


<210>   33
<211>   8
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic

<400>   33

Asp Ile Asn Ser Ala Leu Lys Ser
1               5


<210>   34
<211>   24
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic
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<400> 34

Gly Ala Cys Ala Thr Thr Ala Ala Thr Thr Cys Ala Gly Cys Thr Cys
1   5   10   15

Thr Cys Ala Ala Gly Thr Cys Cys
              20

<210> 35
<211> 8
<212> PRT
<213> Artificial Sequence

<220>
<223> Synthetic

<400> 35

Asp Tyr Asn Ser Ala Leu Ala Ser
1   5

<210> 36
<211> 24
<212> PRT
<213> Artificial Sequence

<220>
<223> Synthetic

<400> 36

Gly Ala Cys Thr Ala Thr Ala Ala Thr Thr Cys Ala Gly Cys Thr Cys
1   5   10   15

Thr Cys Gly Cys Ala Thr Cys Cys
              20

<210> 37
<211> 8
<212> PRT
<213> Artificial Sequence

<220>
<223> Synthetic

<400> 37

Asp Tyr Asn Ser Ala Leu Gln Ser
1   5

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```
<210>  38
<211>  24
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic

<400>  38

Gly Ala Cys Thr Ala Thr Ala Ala Thr Thr Cys Ala Gly Cys Thr Cys
1               5                   10                  15


Thr Cys Cys Ala Ala Thr Cys Cys
            20


<210>  39
<211>  8
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic

<400>  39

Asp Val Asn Ser Ala Leu Gln Ser
1               5


<210>  40
<211>  24
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic

<400>  40

Gly Ala Cys Gly Thr Thr Ala Ala Thr Thr Cys Ala Gly Cys Thr Cys
1               5                   10                  15


Thr Cys Cys Ala Gly Thr Cys Cys
            20


<210>  41
<211>  8
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic

<400>  41

Asp Val Asn Ser Ala Leu Lys Ser
1               5
```

```
<210>  42
<211>  24
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic

<400>  42

Gly Ala Cys Gly Thr Thr Ala Ala Thr Thr Cys Ala Gly Cys Thr Cys
1               5               10              15


Thr Cys Ala Ala Gly Thr Cys Cys
            20


<210>  43
<211>  14
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic

<400>  43

Asp Pro Ala Asp Tyr Gly Asn Tyr Asn Tyr Ala Leu Asp Tyr
1               5               10


<210>  44
<211>  42
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic

<400>  44

Gly Ala Cys Cys Cys Ala Gly Cys Cys Gly Ala Cys Thr Ala Thr Gly
1               5               10              15


Gly Thr Ala Ala Cys Thr Ala Cys Ala Ala Thr Thr Ala Thr Gly Cys
            20              25              30


Thr Thr Thr Gly Gly Ala Cys Thr Ala Cys
            35              40


<210>  45
<211>  14
<212>  PRT
<213>  Artificial Sequence

<220>
<223>  Synthetic
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-15-

<400> 45

Asp Trp Ala Asp Tyr Gly Asn Tyr Asn Tyr Ala Leu Asp Tyr
1   5   10

<210> 46
<211> 42
<212> PRT
<213> Artificial Sequence

<220>
<223> Synthetic

<400> 46

Gly Ala Cys Thr Gly Gly Gly Cys Cys Gly Ala Cys Thr Ala Thr Gly
1   5   10   15

Gly Thr Ala Ala Cys Thr Ala Cys Ala Ala Thr Thr Ala Thr Gly Cys
            20   25   30

Thr Thr Thr Gly Gly Ala Cys Thr Ala Cys
            35   40

<210> 47
<211> 14
<212> PRT
<213> Artificial Sequence

<220>
<223> Synthetic

<400> 47

Asp Pro Ala Asp Tyr Gly Asn Tyr Asp Tyr Lys Leu Asp Tyr
1   5   10

<210> 48
<211> 42
<212> PRT
<213> Artificial Sequence

<220>
<223> Synthetic

<400> 48

Gly Ala Cys Cys Cys Ala Gly Cys Cys Gly Ala Cys Thr Ala Thr Gly
1   5   10   15

Gly Thr Ala Ala Cys Thr Ala Cys Gly Ala Thr Thr Ala Thr Ala Ala
            20   25   30

Ala Thr Thr Gly Gly Ala Cys Thr Ala Cys
            35   40

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```
<210>   49
<211>   14
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic

<400>   49

Asp Trp Ala Asp Tyr Gly Asn Tyr Asp Tyr Ala Leu Asp Tyr
1                   5                   10


<210>   50
<211>   42
<212>   PRT
<213>   Artificial Sequence

<220>
<223>   Synthetic

<400>   50

Gly Ala Cys Thr Gly Gly Gly Cys Cys Gly Ala Cys Thr Ala Thr Gly
1                   5                   10                  15


Gly Thr Ala Ala Cys Thr Ala Cys Gly Ala Cys Thr Ala Thr Gly Cys
                20                  25                  30


Thr Thr Thr Gly Gly Ala Cys Thr Ala Cys
                35                  40
```